

Phytochemicals, Quality and Glycemic Response Fern Red Herbal (*Stenochlaena palustris*)

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ABSTRACT

Red fern (*Stenochlaena palustris*) is an endemic plant that is widely present in open swamps and in tropical rainforests of Borneo. Fern is used as a vegetable by Dayak tribe and has potential for health. Therefore it is necessary to research the utilization of ferns as functional food with the addition of basil leaves as a natural flavor. The research method was performed using Completely Randomized Design (CRD) with a single factor with 6 treats of adding basil leaves of 5 g, 4 g, 3 g, 2 g, 1 g and as a 0 g control. The results of phytochemical analysis on fresh red fern positively contain alkaloids, flavonoids, hydroquinone phenols, steroids, and saponins. The addition of basil leaves has no significant effect on water content, ash content, hedonic and hedonic quality of color, hedonic and hedonic quality of aroma, hedonic and hedonic taste quality, color formation speed without stirring, color forming speed with stirring and color intensity. The best treatment is the addition of basil leaves as much as 5 g with water content of $16.37 \pm 4.88\%$, ash content of $0.95 \pm 0.004\%$, hedonic color like, hedonic quality of brownish red color, aroma rather like, hedonic quality basil-scented aroma, hedonic taste rather like, Hedonic taste very sweet, color formation speed without stirring 7.18 ± 0.75 minutes, color formation speed with stirring 24.85 ± 4.37 seconds, color intensity 0.76 ± 0.05 . EC₅₀ red fern herbal drink is 174,95 ppm. The healthy volunteers who drank a 150 ml red herbal fern drink with the addition of 10 g sugar had a lower glycemic response compared with a control drinking only 150 ml of water with 10 g of sugar added.

Keywords: Red Fern, Phytochemicals, Glycemic Response

I. INTRODUCTION

Increasing the public's attention to good health and food consumption will improve the utilization of functional food sources especially East Kalimantan local food utilization such as herbs onion tiwai based on research result Saragih *et al.* (2014^b) can lower cholesterol patients with hypercholesterolemia by drinking twice Daily in the morning and afternoon. Tiwai can also be used as food additives as food coloring (Saragih *et al.*, 2013; Saragih, *et al.*, 2017). Another plant that is widely used in East Kalimantan is ferns. Red fern (*Stenochlaena palustris*) is an endemic plant that is widely found in open swamps and in tropical rainforests of Borneo. This plant is widely spread in rural areas that have small rivers because the red fern also grows around the banks of the river. This plant has long been exploited by the people of Kalimantan, especially Dayak tribe as vegetables are consumed daily and is a nutritional intake for the Dayak tribe people. Red ferns have many health benefits because they are rich in antioxidants that are good for health.

To increase the useful value of red fern which has many health benefits it needs modern processing innovation that can be used practically and economically by society. One of them is processing red fern as a functional herb. However, the red fern has a disadvantage in terms of the resulting aroma if made as an herbal drink. Therefore, to overcome the problem of the lack of aroma of red fern herbs it is necessary to add a flavor of the plant that has a strong and dominant aroma.

Empirically Indonesian people take advantage of basil (*Ocimum sanctum* L.) to eliminate the unpleasant taste in the mouth, launch breast milk, fever, improve digestion, dilate blood vessels and prevent canker sores. While in China and India, basil is used to maintain health and increase the body's resistance to stress. The content contained in basil leaves are saponins, flavonoids and tannins. The isolated flavonoids in the basil leaf include vicenin, galutenolin, cirsilineol.

Basil leaves have neuroprotective activity, hypoglycemic, antioxidant, antibacterial and anti-inflammatory. Therefore, basil leaves and red fern herbal drinks potentially if used as a functional beverage that is practical to prevent various diseases. This study aims to determine the phytochemical, quality and glycemic response of red fern herbal drinks

II. METHODS AND MATERIALS

Materials

Materials research red fern, basil leaf, alcohol. Tools used such as measuring cylinders, test tubes, analytical scales, furnaces, ovens, desiccators, measuring pipettes, cuvids, spectrophotometers, glucose test kits (Accu Chek Active®).

Research design

This research is a single factor research which is arranged in Completely Randomized Design (CRD) with treatment of addition of basil leaves in red fern herbal, with 6 treatments and 3 replications. Treatment is the addition of natural flavor of basil leaves on red fern B0 = 0: 5 (basil: red fern) B1 = 1: 5 (basil: red fern) B2 = 2: 5 (basil: red fern) B3 = 3: 5 (basil : Red fern), and B4 = 4: 5 (basil: red fern) B5 = 5: 5 (basil: red fern).

The parameters observed in this research are moisture content, ash content, color formation speed, color intensity, phytochemical test, organoleptic test including hedonic scale test and quality, and glycemic response test. Data from organoleptic test which have been obtained then analyzed by variance and for organoleptic test data is converted from ordinal scale data into interval scale using MSI method (Method of Successive Interval)

Research procedure

The process of making red fern herbal in this research is as follows:

- **Sorting**

The materials used in this research are the leaves of red ferns and basil leaves obtained from Dayak Samarinda market, then sorting by way of uniformity of the material size to be used and the degree of aging of each ingredient. Then do the cleaning on the leaves of red ferns and basil leaves that is to remove the dirt-dirt, after it washed in running water.

- **Slicing**

Slicing is done using a knife tool as a cutter. In this slicing process is an advanced stage of the purge stage is done as a goal to reduce the size of red ferns and basil leaves so it will facilitate and accelerate in the process of drying process of the material.

- **Drying**

The drying process stage is done after the slicing process is done. The drying process step is intended to reduce the water content contained in the material. Drying of basil leaf and red fern is done by oven drying method with temperature around 55 ° C for 16 hours until the material is completely dry.

- **Mixing**

At this stage the dried material is then mixed in accordance with the prescribed treatment ratio.

Analysis

Test analysis performed include: 1) water content, 2) ash content, and 3) phytochemical analysis (AOAC, 2006), 4) Organoleptic test done by first brewing with comparison of red grass fern as much as 3 gram by using hot water 150 ml, The preferred scale on the red fern herbal beverage is assessed on a scale of 1-5 and hedonic quality on a scale of 1-7. 5) Color velocity analysis analysis: This analysis is done by entering as many as 3 grams of herbal bark into the glass after it was added boiling water as much as 150 mL. Then it is calculated manually using a stopwatch when water starts to be added in a glass until the color of the brew does not change anymore. 6) Color intensity analysis using UV spectrophotometer. Color intensity analysis using a UV spectrophotometer begins by using water (aquades) as a blank. The sample is the color of red fern herbal extract. Then the blanks are inserted into a UV spectrophotometer with a wavelength of 580 nm (Saragih, 2011). 7). Antioxidant activity by method DPPH (Molyneux, 2004). 8) Glycemic Response Test, a material for the glycemic response test of organoleptic test of red fern herbs with the best natural flavor or the most desirable and favored by panelists. Before the best red fern herbal samples were given to the subject, the objectives of the study were to be explained, then if they were willing to sign the informed consent. Blood sampling was done on the first day by giving 150 ml of red fern herbs (3 g) with 10 gram sugar concentration to 10 healthy volunteers who had undergone full fasting (except water) during the night (around 20.00 until 08.00 am the next day) Blood Collection was done by hose Time 30 minutes 4 times

(minutes to 0, 30, 60, and 90). Then 3 days after the first blood collection, then the second blood taking with the administration of 150 ml of sugar water with sugar concentration as much as 10 grams of control (Saragih, 2014a). Blood glucose measurement using the Accu Check Active Glucometer tool. Blood samples obtained on the surface of the skin after a small bit of injury by using a special lancet (puncture), are touched on the sensor gap at the end of the test strip that has been attached to the digital detector so that the sample glucose level is read.

III. RESULTS AND DISCUSSION

Phytochemical Red Ferns

The results of phytochemical analysis showed that the positive red fern tested contained alkaloids, hydroquinone phenols, flavonoids, saponins, and steroids (Table 1)

Table 1. Phytochemical content of red ferns

	Phytochemicals				
	Alkaloids	Hydroquinone Phenol	Flavonoids	Saponins	Steroids
Red fern	+	+	+	+	+

Description: + = positive

Water Content

Table 1. Water and ash content fern red herbal

Treatment	Water content (%)	Ash content (%)
B0	13.67±0.81	0.93±0.02
B1	13.72±0.33	0.94±0.01
B2	13.87±1.32	0.94±0.02
B3	14.06±1.32	0.95±0.01
B4	14.59±1.89	0.95±0.02
B5	16.37±4.88	0.95±0.01

Based on the results of variance, showed that the addition of basil leaves had no significant effect on the resulting red fern water content. The highest red herbal fern water content produced is about 13.67 ± 0.81% and the lowest is about 16.37 ± 4.88% (Table 1). Red fern herbs with the addition of basil leaves still have a high enough water content allegedly because the components of water in the fern herbal is a component of water that is physically bound so that

the water component inside the red fern is difficult to evaporate. Water content is one characteristic that plays an important role in the composition of food and also contribute to the stability of the quality of a product. The determination of moisture content is useful for expressing the content of the substances in plants as percent dry matter (Abraham *et al.*, 2014). The water content of a sample is closely related to the quality of the sample and the speed of the sample damage, whether microbiological or chemical.

Ash Content

Based on the results of variance, it is known that the addition of basil leaves have no significant effect on the content of red fern ash produced. The resulting content of red fern ash is in the range of 0.93 ± 0.01% to 0.95 ± 0.02% (Table 1). It proves that minerals contained a little and still meet the quality requirements set by the green herbal SNI 01-4453-1998 is a maximum of 8%. Research Anggraini *et al.* (2014) showed that the content of ash produced in herbaceous leaf raw material of pegagan was 13.22%, the result obtained was higher than the ash content produced by red fern herbs.

Color Formation Speed

The results of variance indicated that the velocity of the treatment time without treatment of stirring, or with stirring showed no significant effect on red fern herbs (Table 2). The highest result was control treatment (no addition of basil leaves) that was 5.94 minutes and the lowest result was on the addition of 5 grams of basil leaf that was 7.18 minutes. From the results obtained it is stated that red fern herbs without the addition of basil leaves are able to form a faster color than the addition of 5 grams of basil leaves with treatment without stirring. This is because the red fern has anthocyanin and beta carotene compounds.

Table 2. Time brewing the formation of color and colors intensity of red fern herbal

Treatment	Color formation brewing time without stirring (Minutes)	Color formation brewing time with stirring (Seconds)	Color intensity (Absorbance value)
B0	5.94±0.58	20.82±0.68	0.67±0.09
B1	5.94±0.46	21.61±0.25	0.70±0.01
B2	6.37±0.62	23.37±2.21	0.72±0.11
B3	7.04±0.67	23.61±0.96	0.72±0.06

B4	7.15±0.38	24.71±1.61	0.76±0.07
B5	7.18±0.75	24.85±4.37	0.80±0.05

The time of color formation with the highest agitation resulted from the control treatment was 20.82 seconds and the lowest value was obtained from the treatment of 5 grams of basil leaf that was 24.85 seconds. Stirring treatment plays a role in the rapid process of color formation resulting from the components of basil leaves and ferns will dissolve and break down the enzyme polifenol oxidase.

Color Intensity

The result of variance analysis showed that absorbance value in color intensity analysis by using UV spectrophotometer from each treatment had no significant effect on red fern herbs. The result of absorbance value on the intensity analysis of red fern herbal color is presented in table 2. The results showed that the red fern herb with the addition of basil leaves of 5 g had a higher absorbance value of about 0.80 nm and the red fern herb on the control treatment had a lower absorbance value of about 0.67 nm. It proves that the color produced red ferns and basil leaves have a strong brownish yellow color to absorb light. While the herbal fern red control does not produce a strong yellow absorption. Another study conducted by Saragih (2011) shows that the higher the concentration of the tiwai onion, the higher the intensity of the color.

Color Hedonic Scale and Hedonic Quality

The result of color hedonic scale and hedonic quality of red fern herbal is shown in Figure 1

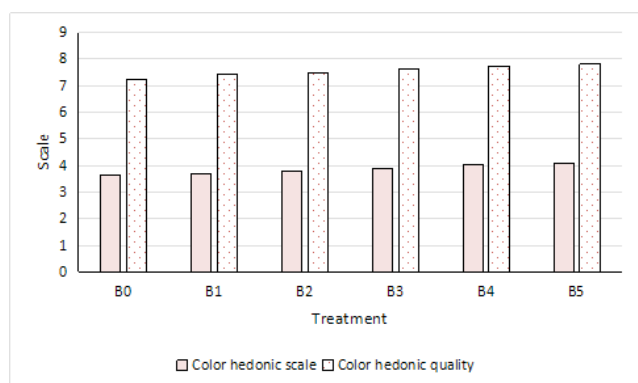


Figure 1. The color hedonic scale and hedonic quality of red fern herbal

Description :

Colors hedonic scale

1 : Very dislike , 2 : Dislike, 3 : Rather like , 4 : Like., 5 : Very like

Colors hedonic quality:

7: Red brown, 6: Rather brownish red, 5: Red gold, 4: Rather red gold, 3: Golden Yellow, 2: Rather yellow, 1: White

The results of variance, showed that the addition of basil leaves have no effect on the color hedonic and hedonic quality of red fern beverage produced. Hedonic color of the highest treatment on addition of 5 grams of basil leaves with a score of 4.09 ± 0.03 (like) and lowest on the control treatment with a score of 3.65 ± 0.12 (rather like). As for the highest hedonic quality of colors in the treatment of 5 grams of basil leaf with a score of 7.82 ± 0.38 (brownish red) and the lowest on the control treatment with a score of 7.24 ± 0.55 (brownish red). In the hedonic quality test panelists prefer the resulting color of red ferns red brownish ferns. Color is a physical parameter formed when light is about an object and is reflected about the sense of sight (eye).

Aroma Hedonic Scale and Hedonic Quality

The results of variance, showed that the addition of basil leaves had no significant effect on hedonic aroma and hedonic quality of red fern herbal aroma. The highest scores of hedonic aroma were obtained on 5 grams of basil leaf addition treatment (3.99 ± 0.14) and the lowest aroma hedonik score was 3.71 ± 0.16 (raher like). As for the highest scores of hedonic aroma quality, the addition of 5 grams of basil leaves is 6.16 ± 0.63 (very strong beroma basil) and for the lowest hedonic quality score is in the control of 5.55 ± 0.27 (aroma fern) as presented in Figure 2.

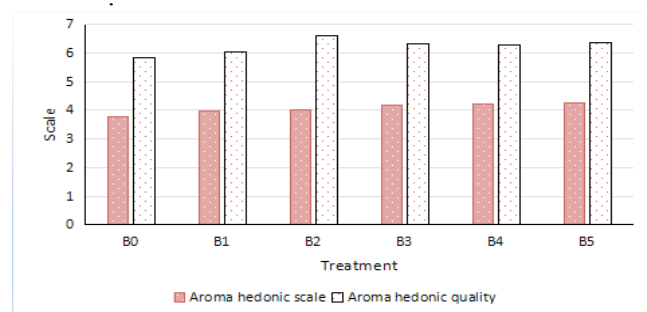


Figure 2. The aroma hedonic scale and aroma quality hedonic of herbal red fern

Description :

Aroma scale hedonic

1 : Very dislike , 2 : Dislike, 3 : Rather like , 4 : Like., 5 : Very like

Aroma hedonic quality:

7: Extremely strongly aroma basil, 6: Very strong aroma basil, 5: Aroma fern
4: Fairly aroma a fern, 3: Rather aroma a fern, 2: Aroma ferns, 1: Very aroma fern

Most of the panelists rather like the scent of basil leaves and red ferns that are less savory or langu. This is in accordance with Lee *et al.* (2013) that the unpleasant aroma of the leaves is in the form of aroma derived from the aliphatic aldehyde compound group of 3-methyl-butanal volatile compounds

Taste Hedonic Scale and Hedonic Quality

The results of variance showed that the addition of basil leaves had no significant effect on hedonic taste and hedonic quality of red fern herbal taste (Figure 3). The highest taste hedonic score was obtained in 5 grams of basil leaf addition treatment which was 4.26 ± 0.19 (slightly like) and the lowest was treated in the control treatment that was 3.76 ± 0.60 (rather like). Then for the highest taste hedonic quality score found in the treatment of 5 grams of basil leaf is 6.61 ± 0.23 (very sweet) and the lowest is in the control treatment is 5.83 ± 0.42 (sweet)

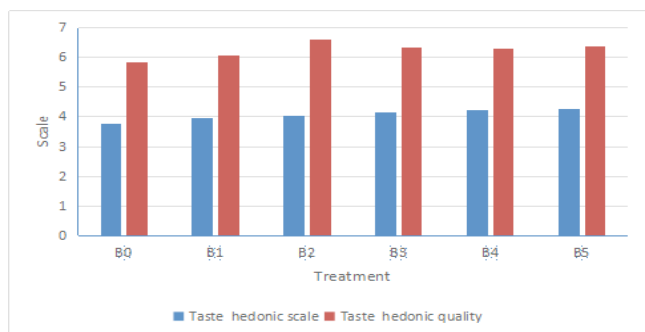


Figure 3. The taste hedonic scale and aroma quality hedonic of herbal red fern

Description :

Taste scale hedonic

1 : Very dislike ,2 : Dislike, 3 : Rather like , 4 : Like., 5 : Very like

Taste hedonic quality:

7: Extremely sweet, 6: Very sweet, 5: Sweet, 4: Rather sweet, 3: Not sweet, 2: Strongly not sweet, 1: Very strongly not Sweet

The best treatment was obtained at the addition of 5 g of basil leaves, and then used for antioxidant activity and glycemic response analysis.

Antioxidant Activity

The antioxidant activity of red fern extract is presented in Table 3. The percentage of DPPH inhibition increased with increasing concentration of red fern extract, at 500ppm inhibition 84.07% (Table 3). The EC50 value calculation is performed by interpolating the percentage of inhibition (50%) into the concentration test solution concentration curve with percentage of inhibition. From data of extract concentration of red fern with inhibition obtained by regression line equation $Y = 0,107X + 31,28$ with value of $EC = 174,95$. Based on the results of this study free antiradical DPPH herbal extract of red fern obtained effective concentration value (EC) 50 of 174.95 ppm. Herbal fern red potential as an antioxidant herbal drink

Table 3. Antioxidant activity of red fern herbal

Concentration (ppm)	Absorbance	% Inhibition
500	0,107	84,07
250	0,272	59,52
125	0,368	45,23
62,5	0,405	39,73
31,25	0,458	31,84

Glycemic Response

The result of comparison of blood glucose response after drinking herbal extract of red ferns plus 10 g sugar and 150 ml of water plus sugar 10 g of blood of respondents are presented in the following Table 4 and Figure 4. Based on the Multiple T-Test test that the red fern herbs and 10 g / 150 mL sugar water were not significantly different. Based on the results in Figure 4 the mean value of the blood glucose response of the subjects to red fern herbs with a sugar concentration of 10 g, 150 mL of water with a sugar concentration of 10 g had a different increase increment from each other.

Average fasting blood glucose volunteers 0 minutes before consuming red fern herbs with a sugar concentration of 10 g is 89.8 ± 10.3 mg / dL, then at minute 30 after drinking herbal fern red blood sugar volunteers have an increase of about $105,1 \pm 12.3$ mg / dL with a difference in blood sugar increase of 14.4 mg / dL. Furthermore, for the 90th minute after consuming the herbal fern red, volunteer blood sugar decreased to about 90.7 ± 8.4 mg / dL with a decrease

of about 5.9 mg / dL. In the 120th minute the blood sugar of the volunteers decreased and returned to fasting glucose which is about 84.8 ± 8.6 mg / dL with a difference of 20.3 mg / dL.

Table 4. Glucose response of red fern herbal and water add 10 sugar (mg/dL)

Healthy volunteers	Red fern herbal water extract 150 ml add sugar 10 g				Water 150 ml add sugar 10 g			
	Minute				Minute			
	0	30	90	120	0	30	60	90
Glucose (mg/dL)								
1	104	108	101	97	87	102	74	79
2	83	105	90	88	102	114	98	90
3	86	125	95	77	86	109	81	86
4	100	113	77	82	94	107	84	84
5	81	103	88	78	85	102	93	88
6	97	95	99	95	90	108	90	90
7	78	87	85	77	87	111	87	78
Average	89.8	105.1	90.7	84.8	90.1	107.	86.	85
Standard WHO				72-126		6	7	.0

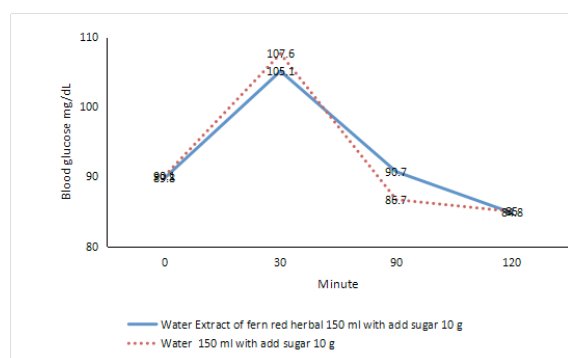


Figure 4. Glucose response of red fern herbal and water add 10 sugar (mg/dL)

The food glycemic index is influenced by several factors, such as the type of materials, processing and characteristics (composition and biochemical properties) of food. The same type of food, when processed in different ways, can have different glycemic indexes (Saragih *et al*, 2014^b; Saragih *et al*, 2017).

CONCLUSION

The addition of basil leaves to the red fern herbs has no significant effect on moisture content, ash content, color formation speed without stirring, color formation speed with stirring, color intensity,

organoleptic analysis including hedonic test and color hedonic quality, hedonic test and hedonic quality of aroma and test Hedonic and hedonic taste qualities. The best treatment was obtained at the addition of 5 g of basil leaves and used for glycemic response analysis. EC₅₀ red fern herbal drink is 174,95 ppm. The healthy volunteers who drank a 150 ml red herbal fern drink with the addition of 10 g sugar had a lower glycemic response compared with a control drinking only 150 ml of water with 10 g of sugar added.

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